AECV91-R4

Interpretation of Ultimate Biochemical Oxygen Demand Data Via Kinetic Curve Extrapolation Models

Alberta

ENVIRONMENTAL CENTRE

INTERPRETATION OF ULTIMATE BIOCHEMICAL OXYGEN DEMAND DATA VIA KINETIC CURVE EXTRAPOLATION MODELS

by

F. Skinner

and

A. van Roodselaar

Environmental Technology Division
Alberta Environmental Centre
VEGREVILLE, Alberta

November 8, 1990

This report may be cited as:

Alberta Environmental Centre. 1990. Interpretation of Ultimate Biochemical Oxygen Demand Data Via Kinetic Curve Extrapolation Models. AECV91-R4

ISBN: 0-7732-0650-7

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

In this report, we explain the methodology and limitations in the interpretation of Biochemical Oxygen Demand (BOD). Databases of up to 257 days are employed to yield Ultimate BOD (UBOD) values. Clear separation of the contribution of carbonaceous and nitrogenous BOD is found to be crucial, and procedures to accomplish this are illustrated. Carbonaceous data sets are processed by a non-linear regression program employing either a one, two, or three term first order model. The two term first order model is definitively superior to the one term model with the three term model showing no significant improvement over the two term model. In physical chemical terms, the two term model corresponds to concurrent fast and slow reaction rates.

Subsets of the data reflecting shorter duration experiments are also analyzed. The UBOD values for the two term model remain fairly consistent until the program fails to converge on a UBOD end value. In contrast, the one term model yields UBOD values that fall farther and farther below the best result as shorter data bases are employed. Statistically, the one term model appears to improve at shorter durations, but this is due to a loss of information of the long term characteristics of the BOD curve.

A review of the ratio of the fast and slow rate of the two term model is carried out for this and other studies. The fast rate ranged from 4 to 28 times the slow rate. In order to evaluate the sensitivity of UBOD to this ratio of rates, non-linear regression is carried out with the fast rate preset to 5, 10 and 20 times the slower. Total UBOD is found to be very insensitive to these variations in the rates. Re-evaluation of non-convergent data sets with restrictions on the rate constants yields UBOD values consistent with other related samples.

PROJECT TEAM

A. Bonneau	L. Peleshok
L. Chrapko	F. Skinner*
S. Davies	P. Truong
H. Hill	A. van Roodselaar*
K. Klingbeil	D. Woznow
A. McFayden	

* To whom correspondence should be addressed

ACKNOWLEDGEMENTS

We would like to thank Standards and Approvals Division, Alberta Department of Environment, for their co-participation in this project. We would like to thank I. McKenzie, L. Noton and P. Carey for the assistance they gave. We would like to thank L. Goodwin for the assistance and expertise he provided during many helpful discussions during the planning and execution of this project. We would also like to thank Z. Florence and T. Kazmierczak for their review of, and assistance in, statistical methodology.

1.0 INTRODUCTION

Most wastewaters contain organic constituents which can be consumed by microorganisms. The degradation of organic matter by aerobic microorganisms will consume oxygen and produce carbon dioxide, water and nitrate (Equation 1):

$$\textit{Organics}\ (C, H, O, N) + O_2 \rightarrow CO_2 + H_2O + NO_3^- \qquad (1)$$

The amount of oxygen required for the oxidation of the organic matter by microorganisms is termed the Biochemical Oxygen Demand or BOD, usually measured in mg/L oxygen.

The BOD procedure in widespread use today employs a 5 day incubation time (1), although reported incubation times have ranged from 15 minutes, as recommended for Immediate BOD (I-BOD) in earlier edition of Standard Methods (2) through 20 days (3), 200 days (4) and 370 days (5). Interest has shifted from the short to the long term procedures. This is in part because of the recognition that the impact of a waste entering the environment will continue at least until all biochemically degradable substances have been consumed. Thus a logical extension of the long term BOD (LT-BOD) is determination of an Ultimate BOD (UBOD). Such a value would be derived either from detection of a plateau in oxygen consumption, or an extrapolation of available trends to a limiting value. Here, the term LT-BOD will be used to refer to the experimental aspects of an extended BOD test, whereas UBOD will refer to the extrapolation of data to an ultimate value.

1.1 Explanation of Procedure

The 5-day BOD procedure is explained in detail in Standard Methods (1). Certain alterations to this procedure are necessary to meet the requirements of the LT-BOD test that may have 50 times the duration. The main differences between the 5-day BOD and the LT-BOD procedure (6, 7, 8) will be highlighted here.

The 5-day procedure recommends dilution of the sample to the point that the consumption of dissolved oxygen (DO) is between about 2 and 6 mg/L during the 5 day experiment, or between 0.2 and 1 mg/L per day. The recommended initial DO consumption for the LT-BOD procedure is at least 1 mg/L per day, but no higher than 6 mg/L per day, as an excessive number of reaerations of the diluted sample would be required. Thus the LT-BOD procedure can employ a higher BOD sample.

Additional nutrients are added to the system during dilution of the sample into the BOD bottles. The effect of various nutrient addition levels on UBOD values is the subject of another publication (9).

The Standard Methods 5-day BOD procedure (1) calls for an initial and a final DO reading. The LT-BOD procedure recommends daily DO readings for the first 30 days. After 30 days, the DO decline was found to be at most 1 mg/L per day, so DO readings could be reduced to twice a week. Typically, DO analyses could be reduced to weekly after 100 days. The high frequency of analysis must be maintained throughout the first thirty days for two reasons. First, the analysis of the results can involve curve-fitting to a relatively complex mathematical expression to derive UBOD constants. For this purpose, many data points in an area of rapid change in DO levels ensure a greater confidence in the fit.

Secondly, initiation of the process of nitrification, the biochemical conversion of ammonia to nitrate and nitrite with simultaneous consumption of oxygen, typically begins between 15 and 20 days after initiation of the test. The rate of oxygen consumption rises dramatically for a few

days, possibly exceeding the initial high rate of DO depletion normally observed at the start of an experiment. Any reduction of the frequency of DO determinations before the end of nitrification could allow exhaustion of the oxygen in the BOD bottle, leading to anaerobic conditions. After 30 days, nitrification is usually complete in experiments carried out at 20°, and the rate of DO consumption drops to below 1 mg/L per day.

The commercially available 300 mL BOD bottles are recommended for the 5 day BOD test. However 2 L BOD bottles have been recommended in some LT-BOD procedures (6, 7). Our studies (9, 10) were largely conducted with 300 mL bottles primarily because of the unavailability of the larger 2 L bottle.

The primary advantage of the 300 mL BOD bottles is their availability. Their primary disadvantage is the need to employ three or more for each BOD test because of the losses of sample during reaeration, as discussed below.

The primary advantage of the 2 L BOD bottles is the possibility of employing a single bottle per sample for the test. The primary disadvantage until very recently was lack of a commercial supplier of these bottles. They are now available from Wheaton Scientific.

One unique requirement of the LT-BOD procedure is to carry out reaerations of the sample to replenish DO levels. As the DO levels approach 2 mg/L during the course of the test, some or all of the sample must be transferred to another vessel, the sample reaerated with clean air, the bottles refilled, the DO values of the replenished test sample obtained, and the incubation resumed.

During this reaeration process and each DO measurement, small losses of sample solution occur. These can be made up in two ways. Either sufficient distilled water can be added to replace the losses, or marbles can be added to displace volume. While the first is the simplest procedure and is recommended by one procedure (6), the effect of repeated minor dilutions must be considered on the final results. Thus a single sample bottle may be opened for up to 100 DO

readings and 20 reaerations. This could result in losses of between 10 and 50 mLs in a single bottle. This represents between 3 and 15% losses for a 300 mL bottle, or between 0.5 and 2% losses for a 2 L bottle.

It was felt that dilution with distilled water would distort the results of some tests carried out in 300 mL bottles. Therefore the procedure utilized calls for three 300 mL BOD bottles to be employed for each test, with two bottles used for analysis, and the third bottle used to make up the losses. The sample taken from the third bottle was made up with acid-washed marbles. This resembles a previously published procedure (7).

Distilled water was used to make up volume losses in the 2 L bottles. The much smaller effect of dilution on the 2 L bottles, a maximum of 2%, was considered comparable or less than experimental error. No corrections were made to account for the small quantities of water added.

Termination of the test has been recommended when the weekly DO consumption is between 1 and 2% (7), or alternatively less than 1%, of the cumulative DO consumption (6). In some cases, our studies continued well after these criteria were met.

The operational aspects of the standard 5-day and this laboratory's interim long term BOD procedures are noted in Table 1.

TABLE 1

Comparison of LT-BOD and 5-Day BOD Procedural Requirements

Factor	LT-BOD	5-Day BOD
Term	144 - 365 days	5 days
Initial DO Decline	1 - 6 mg/L/day	0.2 - 1 mg/L/day
DO Readings Taken	60 - 100	2
Bottle Size	300 mL or 2 L	300 mL
Reaerations	up to 20	0

The reproducibility of the LT-BOD experimental procedure was evaluated by conducting 39 tests at various conditions in duplicate (a total of 78 experiments), and one test with six replicates (a further six experiments). The six replicates are treated as three duplicate runs in the following discussion. The final BOD's of each pair was compared. The average coefficient of variation (CV) was 3.47%. Of the 42 duplicates, 38 fell within 2 CV and 2 fell between 2 and 4 CV. Two of the 42 duplicates showed 25 - 30% variation in the final values. The standard error of the mean CV of all 42 was 0.91, whereas removal of the outliers yielded a standard error of 0.38.

In excess of 60 LT-BOD tests were carried out at different temperatures and concentrations, employing treated effluent from several pulp mills. This paper will report a portion of the results obtained at 20°, focusing on the interpretation of the data of a limited set of analyses.

2.0 INTERPRETATION OF RESULTS

2.1 Scenarios for BOD Change

The simplest interpretation of BOD data would be based on the analogy of a single micro-organism supplied with one compound as a carbon growth substrate. The growth rate of the organism is dependent solely on the concentration of that compound (11). The population of the organism will initially increase rapidly, but will eventually reach a limit set by the finite source of carbon and energy available (see Figure 1).

The rate of disappearance of the carbon source as measured by BOD can be expressed as:

$$\frac{dL}{dt} = -kL \qquad (2)$$

where k is the rate coefficient and L is the BOD remaining at time t. At any time t, the UBOD is the sum of BOD remaining and BOD consumed. This can be expressed as:

FIGURE 1:
SIMPLE EXPONENTIAL GROWTH

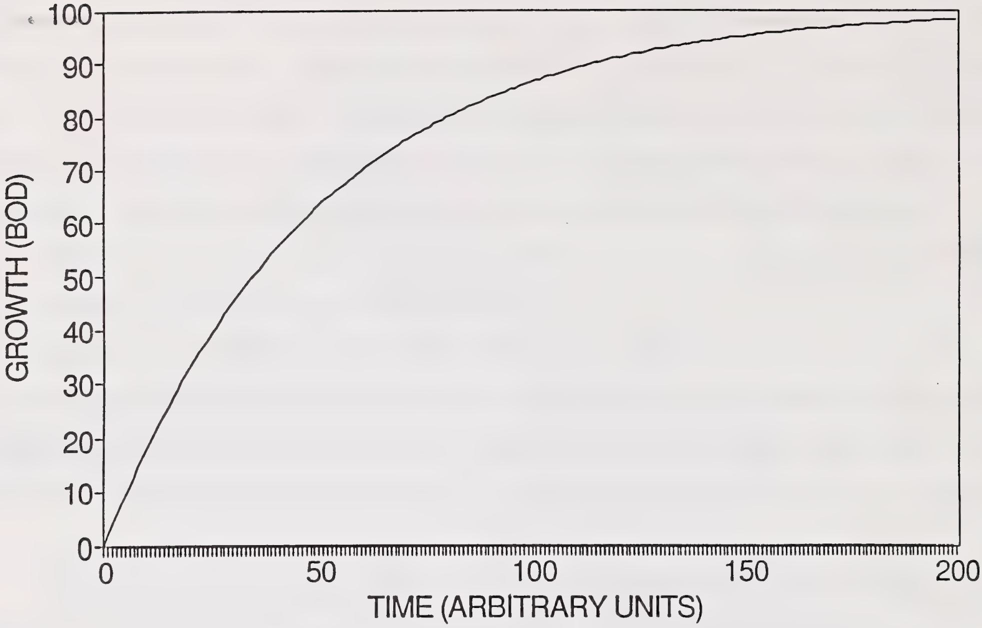
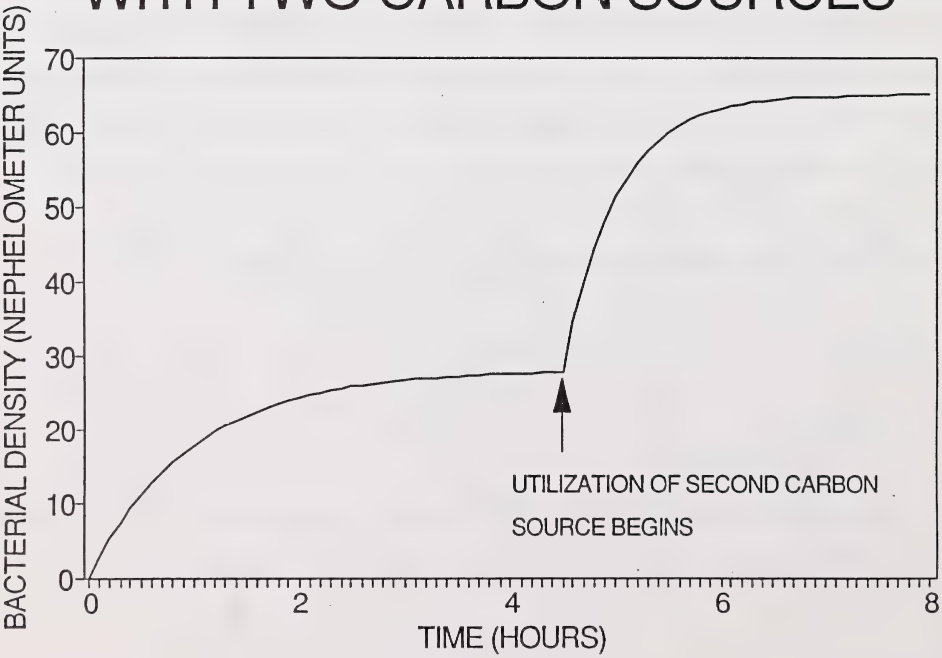


FIGURE 2: SINGLE ORGANISM TYPE WITH TWO CARBON SOURCES



$$L = UBOD - BOD(t) \tag{3}$$

where BOD(t) is the BOD consumed by time t. Integration of Equation 2 and substitution of L in Equation 3 yields:

$$BOD\,(t) = UBOD\,(1 - e^{-kt}) \tag{4}$$

A single organism, when provided with <u>two</u> utilizable carbon growth substrates can exhibit the "diauxy" effect. In such cases the more easily degradable compound is utilized first; growth often slows down or ceases as this substrate is exhausted only to recommence after a delay as the alternate substrate is utilized (12) (See Figure 2). Such a curve, idealized to account for immediate initial metabolism of one carbon source followed by a delay before metabolism of the second carbon source, can be expressed by:

$$BOD(t) = UBOD_1\,(1 - e^{-k_1 t}) + UBOD_2\,(1 - e^{-k_2(t - t_0)}) \tag{5}$$

where $UBOD_2$ and k_2 are magnitude and rate parameters relating to the second carbon source, and t_o is the observed delay before utilization of the second carbon source commences.

The actual situation in a waste sample is far more complex, with many chemicals present in small quantities, and a wide variety of organisms being present in the wastewater sample or introduced in the seed. A thorough treatment of a typical waste would consider all components, each of which may be depleted at a different rate by different types of micro-organisms. The phenomenon of delayed metabolism of some components must be included. This could be expressed as:

$$BOD(t) = \sum_{i=1}^{n} \sum_{x=1}^{m} UBOD_i\,(1 - e^{-k_i t}) + \sum_{j=1}^{p} \sum_{y=1}^{q} UBOD_j\,(1 - e^{-k_j(t - t_{oj})}) \tag{6}$$

where i refers to all chemical constituents that begin metabolism at once, x refers to strains of micro-organism metabolizing each of these constituents, j refers to constituents metabolized after a delay of t_o, and y to strains of micro-organisms utilizing these constituents. Additional complexity such as the contributions of secondary metabolites or synergistic effects are not considered.

Certain simplications can be proposed to this analysis. First, the only significant delayed DO demand in the time scale of the LT BOD analysis is attributable to the effects of nitrification (6, 7, 13, 14). This can be largely accounted for by monitoring a blank, which would have the same nutrients as added to the waste sample. The oxygen demand is attributable to the biochemical conversion of ammonia present in the nutrient solution or derived from organic amines to nitrate (7):

$$NH_4^+ + 2O_2 \rightarrow NO_3^- + 2H^+ + H_2O \tag{7}$$

This reaction consumes 4.57 mg of oxygen per mg of ammonia nitrogen.

Second, some researchers have proposed that kraft pulp mill effluents exhibit a two stage BOD process with the first rapid stage attributable to the oxidation of carbohydrate constituents and the second slower stage to lignin decomposition (15, 5). Similarly Crawford et al (16) suggested kraft lignin underwent rapid degradation for the first 4 - 8 days of incubation, and degraded slowly thereafter. The identification of the observed processes with lignin degradation is somewhat doubtful since other authorities (17) believe that only a specialized group of microorganisms, the white-rot fungi, can degrade lignin. Bouveng and Solyom (18) also noted a two stage process in a 40 week study. Combination of two stages of BOD removal and the effect of nitrification would yield the following:

$$BOD(t) = UBOD_1 \, (1 - e^{-k_1 t}) + UBOD_2 \, (1 - e^{-k_2 t}) + UBOD_3 \, (1 - e^{-k_3 (t - t_d)}) \tag{8}$$

$UBOD_1$, $UBOD_2$ and $UBOD_3$ refer to rapid, slow and delayed BOD respectively. The first two terms can be attributed to carbonaceous BOD, and the delay term to nitrogenous BOD by this rationale.

R.C. Whittemore and T. Hovis evaluated 9 different regression models for the Long Term BOD analysis of 9 effluents after removal of the effects of nitrification (13, 19). The most satisfactory model for the carbonaceous BOD was found to be the two term expression:

$$BOD(t) = UBOD_1 \, (1 - e^{-k_1 t}) + UBOD_2 \, (1 - e^{-k_2 t}) \tag{9}$$

This two term model has indeed been used in practice by other workers for carbonaceous UBOD analyses (4, 6, 7, 20).

In contrast, there exists a significant body of literature that employs non-linear regression with the single term exponential Equation 3 (21, 22, 23, 24, 25, 26) to evaluate UBOD. However all of these studies employed experimental terms of between 15 and 30 days, far less than the term employed in studies recommending the two term exponential Equation 9.

Hewitt et al (27) evaluated other more complex kinetic models. He concluded that while the statistical quality of the results improved, the UBOD's found were unrealistically high, and "do not represent actual environmental levels".

2.2 Treatment of Data

Because of the desirability of analyzing just the carbonaceous BOD, the effects of nitrification had to be removed from the BOD data. This could be accomplished in a variety of ways. Thus routine analysis of total kjeldahl nitrogen, ammonia, nitrate and possibly nitrite concentrations at the same time as the DO analysis would allow exact corrections based on calculated oxygen requirements for the various oxidative steps. Because of the large scope of our study, this was impractical due to the resultant analytical load.

Secondly, the addition of inhibitor to eliminate the effects of nitrification has been employed (1, 4, 6, 7). However, McKeown et al (4) reported that two nitrification suppression chemicals also inhibited some portion of the carbonaceous oxygen demand. Conversely, other workers (6) found that the inhibitors could become ineffective in 50 to 100 days, and may add to the oxygen demand. This method was therefore not employed.

Thirdly, the nitrification observed in a blank containing identical nutrients could be used to correct the analytical results. This implies that the contribution of the sample to the effects of nitrification is quite small compared to the ammonia contained in the nutrients which are

added to each sample and blank. Indeed, the levels of ammonia in samples analyzed was less than 5% of the nutrient loading. Consequently, this method was adopted in the current study.

The raw data from the blank contained typical experimental scatter as illustrated in Figure 3. A direct subtraction of blank BOD contribution from a sample BOD would accentuate these imperfections, reducing the likelihood of a satisfactory mathematical fit of the curve to any exponential functions. For this reason the blank BOD data was itself analyzed by Fitall, an iterative non-linear regression program (28), which employs least squares fitting via Marquardt's algorithm.

The best delayed exponential curve determined for a typical blank gave a less than ideal fit. However inclusion of a small linear increase term yielded an excellent fit, as illustrated in Figure 4. The curve used is expressed:

$$BOD\ (blank) = L_1\ (1 - e^{-k_1(t - t_d)}) + L_2 t \qquad (10)$$

The physical significance of the constant L_2 is not immediately clear. Young et al (29) illustrated such a linear increase, but did not comment on it.

The initiation of nitrification in BOD studies conducted at 20 degrees typically occurs between 10 and 25 days. The reason for the variation is attributable to a number of causes, such as varying populations of microorganisms, and enhancement or retardation of the growth of nitrifying bacteria by constituents in the sample. In our studies, the initiation time of nitrification of the blank did not necessarily match that of the samples. Thus if simple subtraction of the blank contribution to total BOD were utilized, an apparent dip or alternatively a spike could appear in the corrected data, as illustrated in Figure 5 and 6 respectively. For this reason the BOD data from the blank was visually evaluated with varying delays, and the delay that minimized or eliminated any peak or dip was employed for curve fitting analysis.

After blank subtraction, the blank corrected BOD curves were subjected to non-linear regression analysis using Fitall, an iterative computer program. Initialization parameters were

FIGURE 3
OBSERVED BLANK CURVE

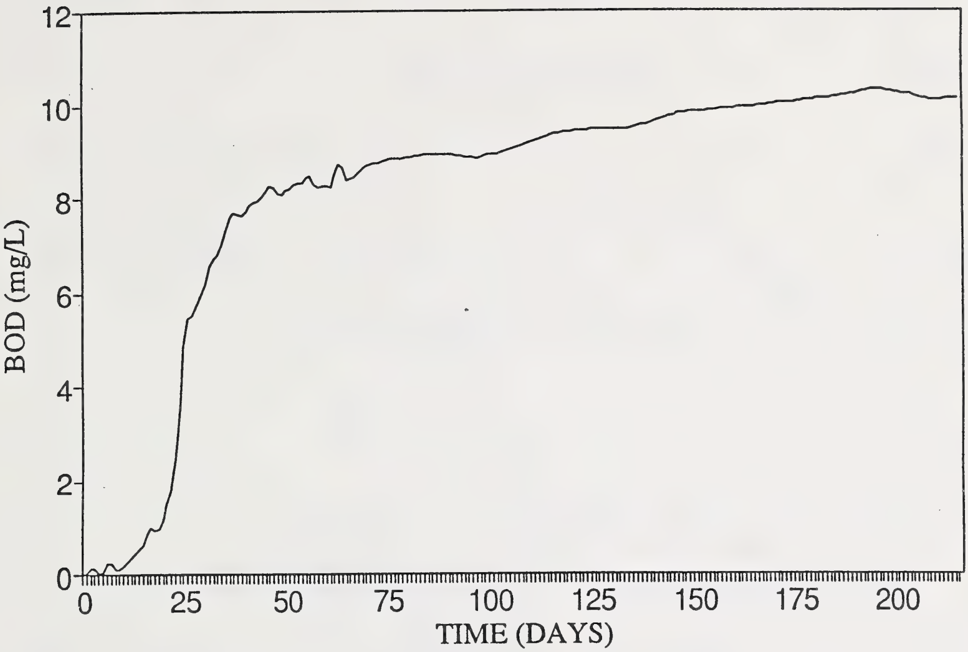


FIGURE 4: OBSERVED BLANK WITH ONE AND TWO TERM FITTED CURVES

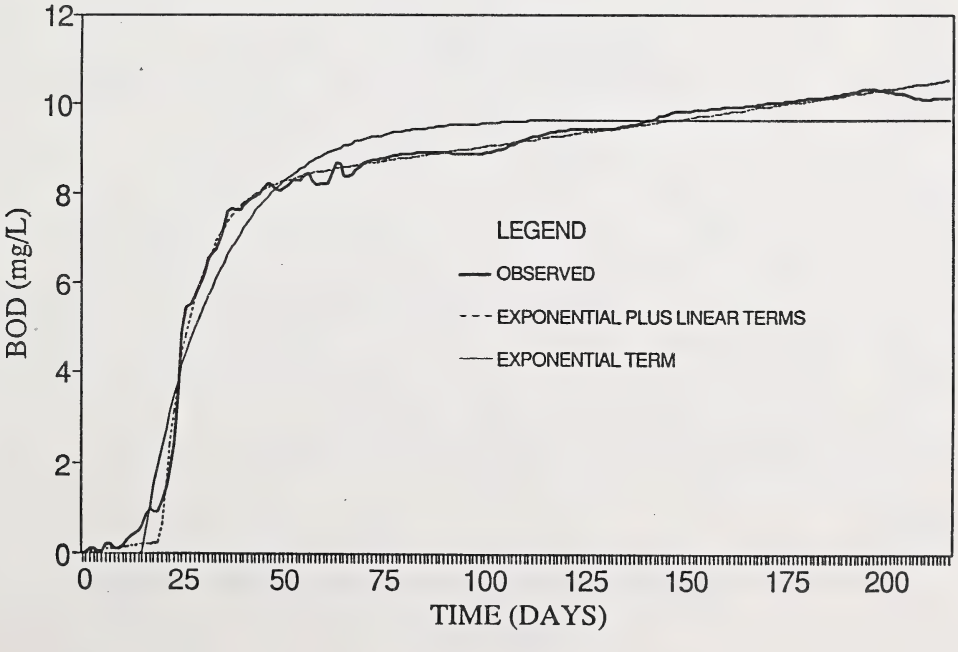


FIGURE 5: BLANK CORRECTION OF SAMPLE 11 USING AN UNDERESTIMATED INITIATION TIME

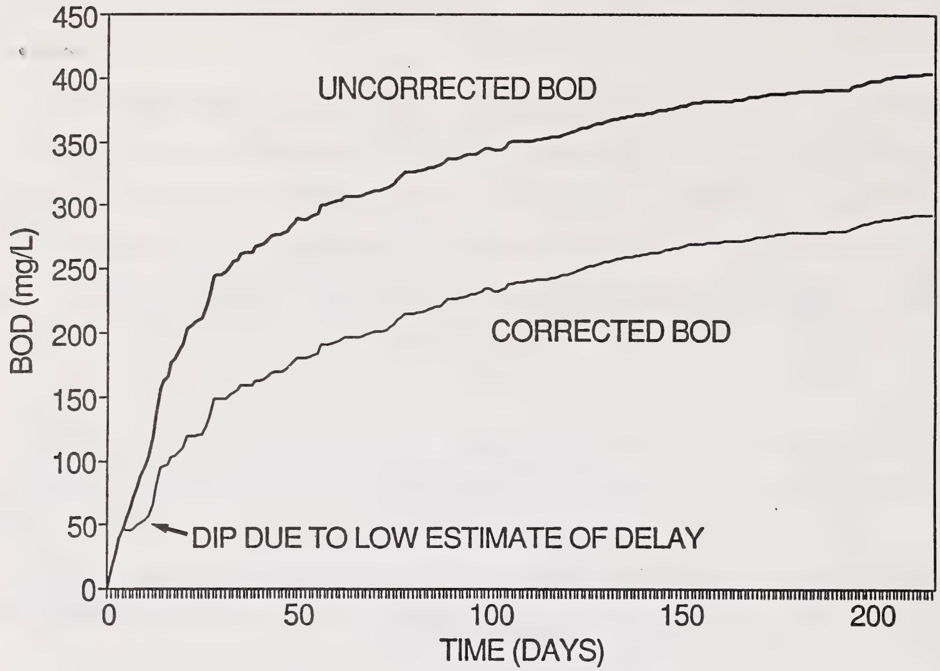
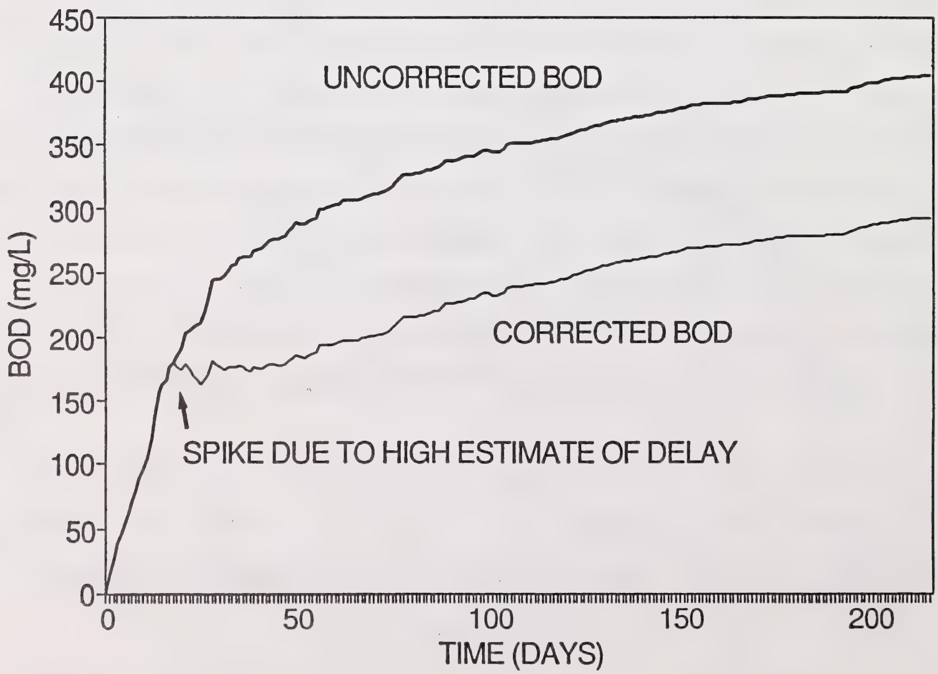


FIGURE 6: BLANK CORRECTION OF SAMPLE 11 USING AN OVERESTIMATED INITIATION TIME



set to intermediate values of UBOD and rate. The most stringent criteria, a change in the variance of less than one part in 10" in successive iterations for termination of the Fitall program were used.

Three expressions were tested:

A. Single exponential

$$BOD\ (t) = UBOD\ (1 - e^{-kt}) \tag{4}$$

B. Two term exponential

$$BOD(t) = UBOD_1\ (1 - e^{-k_1 t}) + UBOD_2\ (1 - e^{-k_2 t}) \tag{9}$$

C. Three term exponential

$$BOD(t) = UBOD_1\ (1 - e^{-k_1 t}) + UBOD_2\ (1 - e^{-k_2 t}) + UBOD_3\ (1 - e^{-k_3 t}) \tag{11}$$

2.3 Non-Linear Regression with the One Term Exponential Function

The results for the single exponential equation 4 are noted in Table 2. These results include effluents from pulp mills using varying processes, with different nutrient levels, and varying effluent concentrations. In spite of the great variety of sample sources and treatments, which yielded UBOD values from 175 to 2466 mg/L, the rate k varied only from 0.0236 to 0.0600. The rate of oxygen depletion is thus a relatively stable parameter in this analysis.

The quality of the results can be measured by the determination index, r^2 (30). This corresponds to the coefficient of determination commonly used in linear regression, and is the square of the correlation coefficient r. Thus

$$\textit{determination index } (r^2) = 1 - \frac{\textit{unexplained sum of squares (USS)}}{\textit{total sum of squares (TSS)}} \tag{12}$$

$$= 1 - \left[\frac{\Sigma\,(Y_{OBS} - Y_{CALC})^2}{\Sigma\,(Y_{OBS} - \overline{Y})^2} \right]$$

TABLE 2

Ultimate Biochemical Oxygen Demand and Rate Constants for Blank-Corrected BOD Results Determined by Non-Linear Regression for Equation 4:

$$BOD(t) = UBOD\,(1 - e^{-kt})$$

Sample	UBOD	k	Determination Index (r^2)
1	285.8	0.0319	0.9954
2	305.1	0.0292	0.9958
3	265.6	0.0349	0.9366
4	342.8	0.0286	0.9723
5	339.6	0.0303	0.9850
6	372.7	0.0284	0.9943
7	275.9	0.0364	0.9588
8	314.8	0.0349	0.9590
9	175.7	0.0265	0.6045
10	240.2	0.0269	0.9008
11	255.5	0.0236	0.9426
12	283.0	0.0247	0.9668
13	2111	0.0378	0.9873
14	2167	0.0374	0.9847
15	2296	0.0600	0.9363
16	2466	0.0498	0.9515

FIGURE 7: OBSERVED AND CALCULATED BODS OF SAMPLE 3 FOR ONE TERM FIT

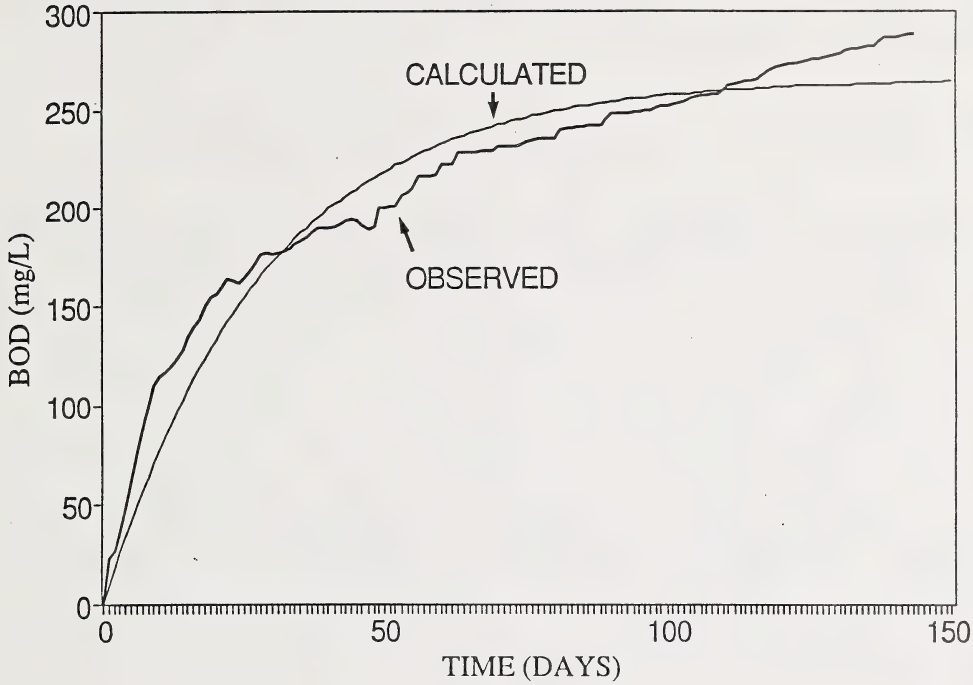
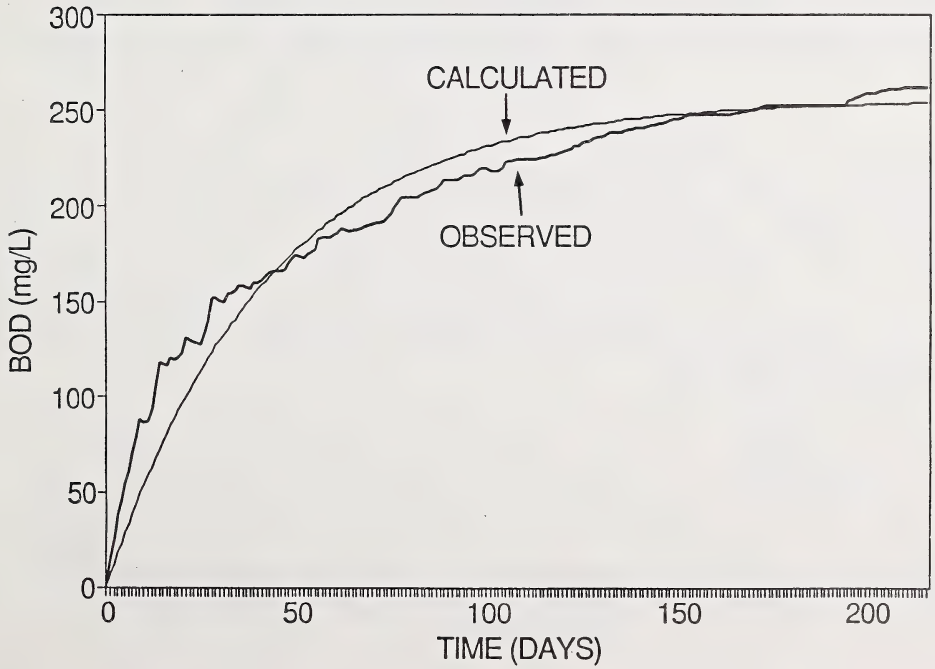


FIGURE 8: OBSERVED AND CALCULATED BODS OF SAMPLE 11 FOR ONE TERM FIT



Half of the regression results have a determination index in excess of 0.96. However two regression results, samples 9 and 10, yield determination indices below 0.92. There is therefore a substantial qualitative difference in results.

Two typical BOD curves and the fitted results are illustrated in Figures 7 and 8.

The source of the unexplained variation could be ascribed to random or systematic differences between the observed and calculated results. One effective means for discerning if there is indeed a pattern to the variation is to plot the difference or residual between these values. Two typical residual plots are illustrated in Figures 9 and 10. In these and other one term residual plots, there are consistent trends, suggesting a different mathematical curve could provide a better result. Noted on Figures 9 and 10 are histograms from a univariate procedure on the residuals. A normally distributed residual would produce a "bell-shaped" normal curve, unlike these histograms. Also noted are the means (T: mean = 0) expected to be 0 and the sum, also expected to be 0 for a normally distributed residual. The significance of these will be discussed with the two term fit results.

2.4 Non-Linear Regression with the Two Term Exponential Function

The data was then fitted to the 2 term exponential function expressed in Equation 9. The results are noted in Table 3. The two BOD curves and the two term fit are shown in Figures 11 and 12.

One point of concern is immediately obvious from these results. For samples 6 through 9, the non-linear regression program did not converge on an answer in 300 iterations, and their UBOD is many times the UBOD noted in the single term non-linear regression noted in Table 2. These results clearly contrast with all results that did successfully converge, and will be treated separately in the discussion.

FIGURE 9: RESIDUALS FOR THE ONE TERM EXPONENTIAL EQUATION FOR SAMPLE 3

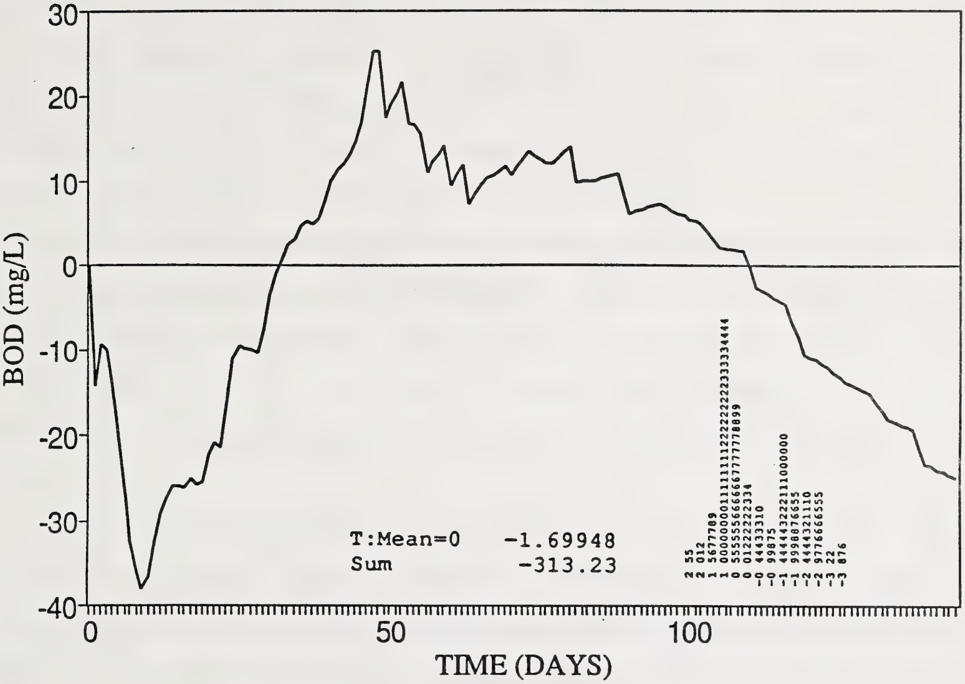


FIGURE 10: RESIDUALS FOR THE ONE TERM EXPONENTIAL EQUATION FOR SAMPLE 11

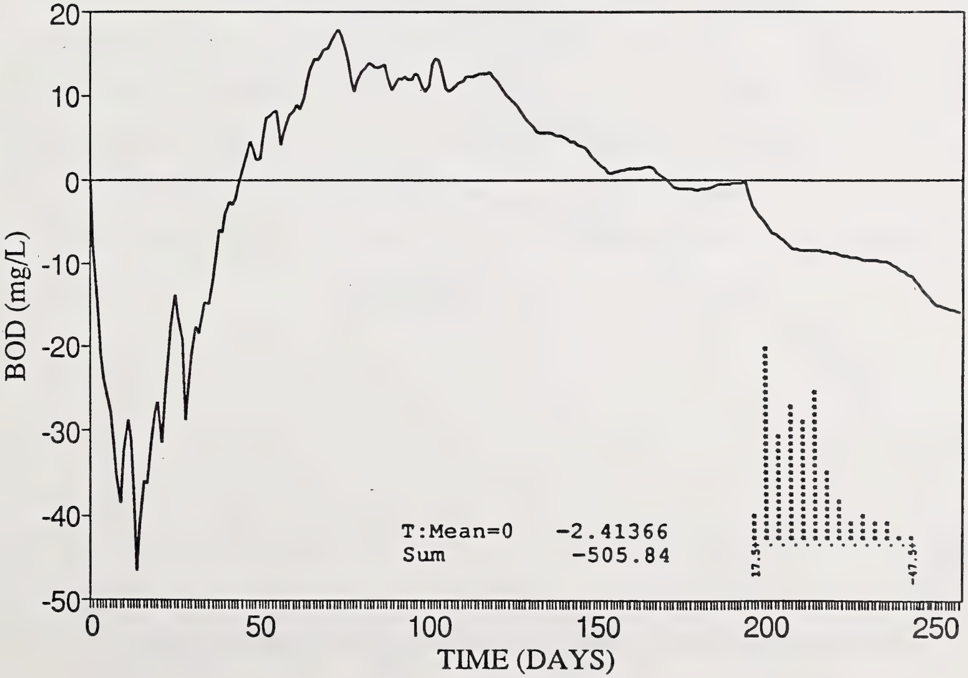


FIGURE 11: OBSERVED AND CALCULATED BODS OF SAMPLE 3 FOR TWO TERM FIT

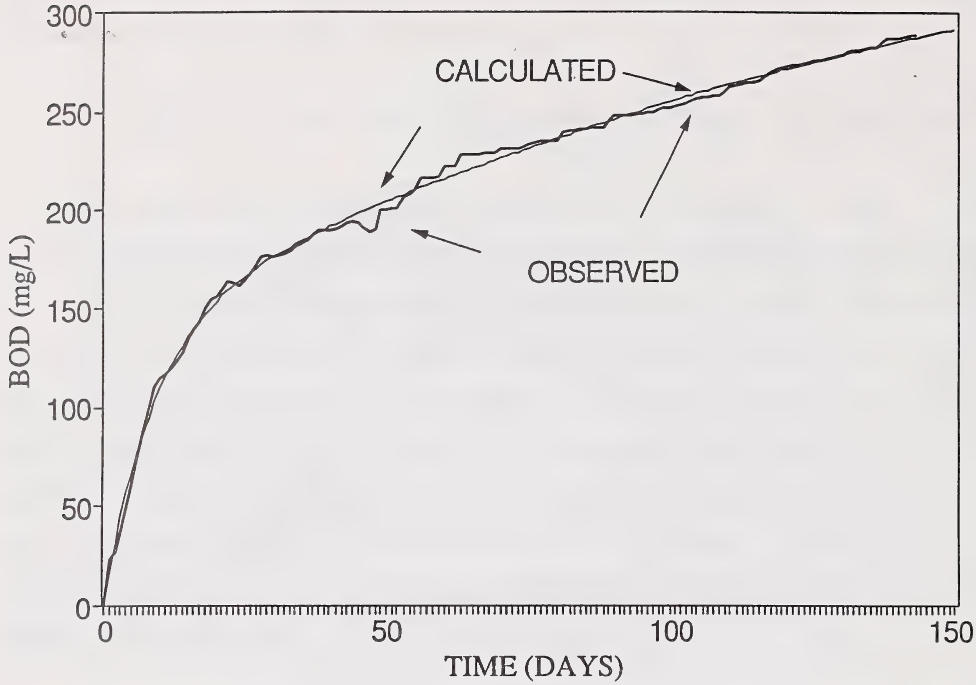
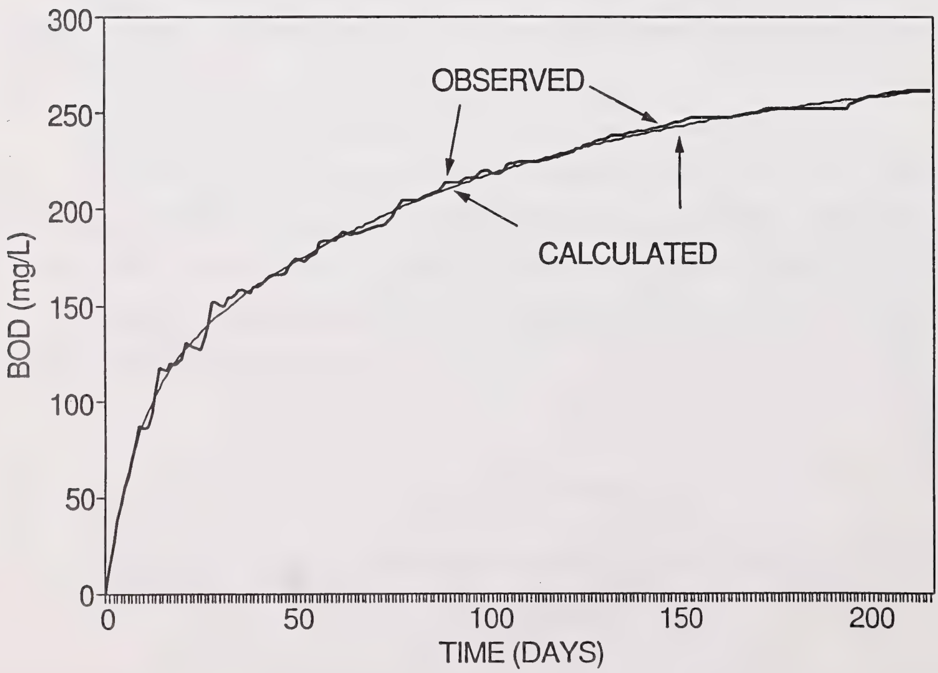


FIGURE 12: OBSERVED AND CALCULATED BODS OF SAMPLE 11 FOR TWO TERM FIT



The total UBOD noted in Table 3 for convergent results was consistently larger than the UBOD determined by the single term curve-fitting exercise noted in Table 2. The determination index was significantly improved, and the residuals showed no obvious trends, as illustrated for two samples in Figures 13 and 14. Statistical analysis helped define if the one term and two term residuals were random and normally distributed or not. Clearly, the histograms of the two term residuals from univariate analysis approximate a bell shaped normal distribution, although only Sample 11 two term residuals were normally distributed at $P > 0.05$, based on S-W test. The mean values of the residuals are expected to be 0 in a normal distribution. Indeed, both two term residual means are not significantly different from 0. However for the one term residuals, sample 3 is marginal at $P = < 0.09$ and sample 11 is significantly different at $P = < 0.02$. Further statistics that favour the two term fits are the values of the "sum", which are expected to be 0.

The four residual plots all plainly demonstrate non-random trends. The notable difference between the two models is that in the one term model both the mean and variance of residuals are trended over time, while the two term model results in stable periodicities, i.e. similar to a sine function. The source of this periodicity may lie in some of the simplifying assumptions necessary to derive the two term exponential under discussion, or in an artifact due to the experimental procedure.

Table 4 gives a comparison of the one-term and two-term results. There is not a consistent ratio between the UBOD values noted in Table 2 and Table 3. Thus the single term derived UBOD was within 10% of two term derived UBOD in seven of the twelve cases, within 20% in three more cases, and over 30% less in two cases. Generally the UBOD's determined by the two methods that most closely match each other have the better correlation indices in Table 2.

FIGURE 13: RESIDUALS FOR THE TWO TERM SOLUTION FOR SAMPLE 3

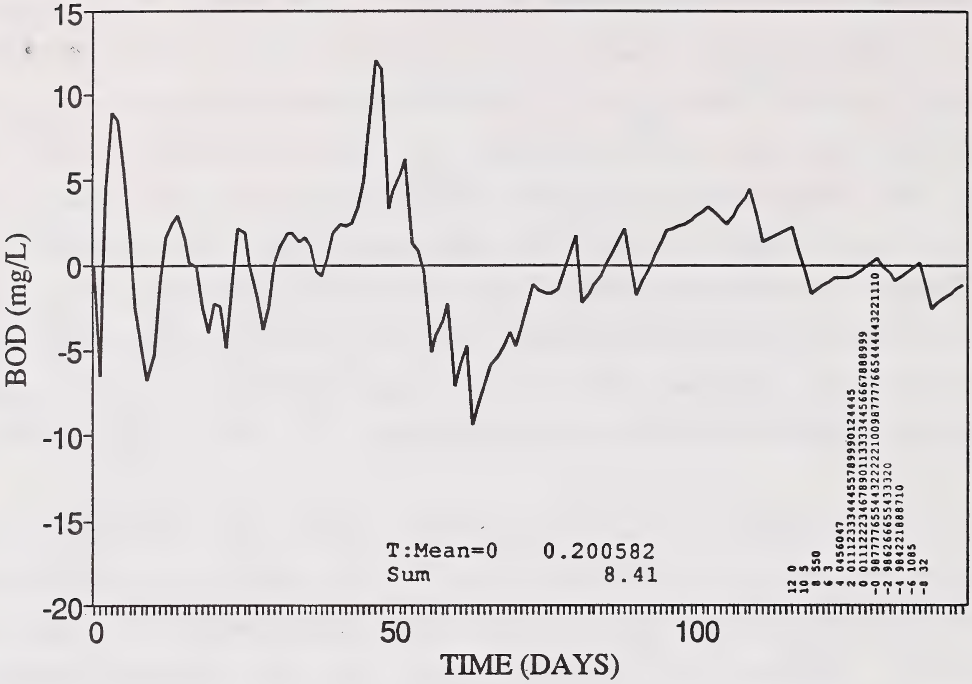


FIGURE 14: RESIDUALS FOR THE TWO TERM SOLUTION OF SAMPLE 11

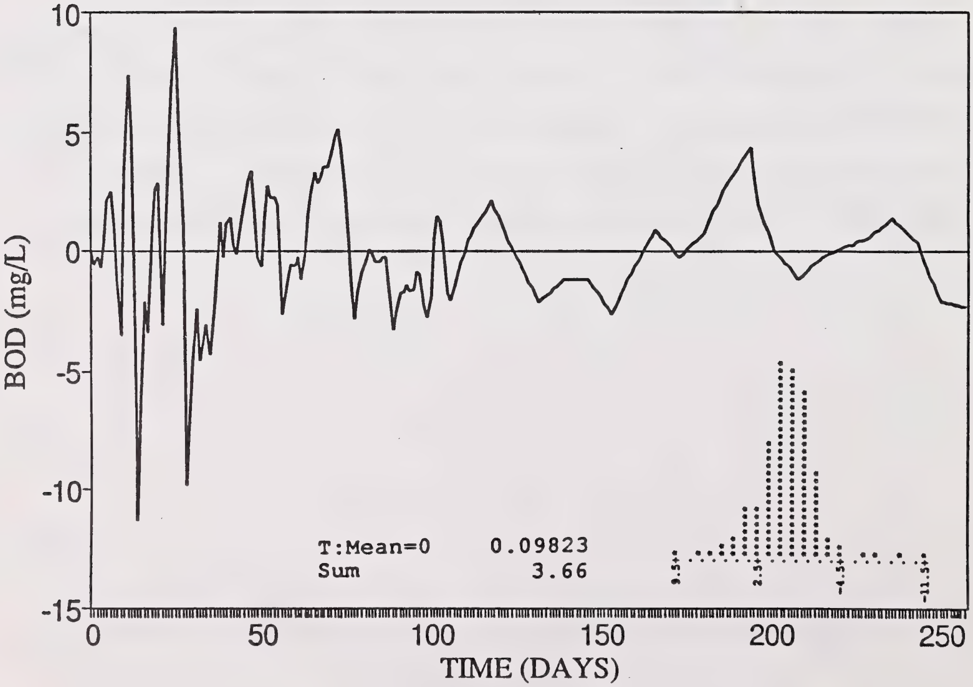


TABLE 3

Ultimate Biochemical Oxygen Demand and Rate Constants for Blank-Corrected BOD Results Determined by Non-Linear Regression with Equation 9:

$$BOD(t) = UBOD_1\,(1 - e^{-k_1 t}) + UBOD_2\,(1 - e^{-k_2(t)}) \qquad (9)$$

Sample	Total UBOD	$UBOD_1$	k_1	$UBOD_2$	k_2	Determination Index (r^2)
1	291	25	0.299	266	0.0279	0.9981
2	310	23	0.312	287	0.0258	0.9981
3	386	136	0.118	250	0.0065	0.9968
4	390	103	0.142	287	0.0142	0.9973
5	401	164	0.068	237	0.0108	0.9974
6*	2870	320	0.034	2550	E-4 1.55	0.9955
7*	5574	192	0.067	5382	E-4 1.37	0.9880
8*	21259	229	0.056	21030	E-5 4.9	0.9775
9*	31390	80	0.252	31310	E-4 2.52	0.8498
10	355	95	0.130	260	0.0065	0.9406
11	279	97	0.130	182	0.0109	0.9982
12	318	153	0.060	165	0.0083	0.9980
13	2154	640	0.126	1514	0.0256	0.9989
14	2229	903	0.096	1326	0.0216	0.9988
15	2395	1473	0.130	922	0.0194	0.9841
16	2575	1407	0.122	1168	0.0193	0.9931

* No convergence; regression program halted after 300 + iterations.

TABLE 4

Comparison of Total UBOD and Correlation Index Determined by Non-Linear Regression Employing Equations 4 and 9.

Sample	UBOD		Ratio of UBOD (One Term / Two Term)	Determination Indices r^2		Ratio of Variances*
	One Term (Table 2)	Two Term (Table 3)		One Term (Table 2)	Two Term (Table 3)	
1	286	291	0.98	0.9954	0.9981	2.5
2	305	310	0.98	0.9958	0.9981	2.3
3	266	386	0.69	0.9366	0.9968	19.4
4	343	390	0.88	0.9723	0.9973	10.3
5	340	401	0.85	0.9850	0.9974	5.8
10	240	355	0.68	0.9008	0.9406	2.1
11	256	279	0.92	0.9426	0.9982	32.1
12	283	318	0.89	0.9668	0.9980	16.6
13	2111	2154	0.98	0.9873	0.9989	12.0
14	2167	2229	0.97	0.9847	0.9988	13.2
15	2296	2395	0.96	0.9363	0.9841	4.0
16	2466	2575	0.96	0.9515	0.9931	6.9

* Ratio of variances is defined as variance (one term)/variance (two term)

In the two worst cases, the two term UBODs were 1.45 and 1.47 times the one term result. We propose that a maximum value for the two term UBOD could be conservatively estimated by taking 1.5 times the UBOD determined by the single term exponential expression. This suggested rule of thumb could be used for experiments of at least 120 days duration, as at shorter terms, the non-linear regression with a single term exponential steadily declines. This will be discussed in detail below.

A better statistic to evaluate the quality of the one and two term fits is based on differences between the variances. As noted in Table 4, the ratio of the variances of the one term fit to the two term fit was always greater than 1. However, while the two term variance is superior in the 12 cases in Table 4, can this be considered a statistically significant difference at, say, the 0.01 level?

Evaluation of the significance was carried out with the F table. Essentially, if the ratio of variances exceeds the value for F at the appropriate number of degrees of freedom at the 0.01 level, then the variances can be considered significantly different. The most conservative value noted in the F table close to the short 144 day trials, $F(100, 125, 0.01) = 1.54$, is smaller than all of the ratios noted in Table 4. Therefore the variance of the two term fit is significantly smaller than the one term fit in all 12 cases.

2.5 Non-Linear Regression with the Three Term Exponential Function

The dramatic improvement in the statistical quality of the two term exponential fit posed the question of whether a three term exponential equation would offer further insight to the biochemical situation. Application of the non-linear regression program with Equation 11, however, yielded either a result very similar to the two term exponential or a non-converging solution in every case except two.

Sample 6 and 13 yielded meaningful UBOD values noted in Table 5 with the three term exponential expression. For sample 6, this contrasts to the two term expression which never converged to a meaningful value. The significance of the very fast term is effectively to reduce the BOD by 12, at which point the slightly modified data fit the normal two-term exponential expression. This very fast component does not have any biochemical meaning, so the very fast UBOD term supplies no further useful information for Sample 6. The determination index shows a minor improvement from 0.9955 in the two term solution to 0.9959, but this is not significant.

The three term solution for Sample 13 shows a 3% increase in UBOD - 2228 compared to 2154 - and the appearance of a very slow term. However the negligible change in the determination index, 0.9992 compared to 0.9989 for the two term expression, gives little reason to consider the differences crucial.

TABLE 5

UBOD Parameter Determined for Blank-Corrected Data from Samples 6 and 13 with Equation 9:

$$BOD(t) = UBOD_1\,(1 - e^{-k_1 t}) + UBOD_2\,(1 - e^{-k_2 t}) + UBOD_3\,(1 - e^{-k_3 t}) \qquad (9)$$

Sample 6	UBOD	k	Determination Index
Total	412	NA	0.9959
Very fast	12.0	7.08	NA
Slow	306	0.0312	NA
Very Slow	94.2	0.0065	NA
Sample 13	UBOD	k	Determination Index
Total	2228	NA	0.9992
Fast	386.6	0.1932	NA
Slow	1539	0.0344	NA
Very Slow	302.6	0.0072	NA

As only two of sixteen samples gave any successful three term solutions, and those two showed negligible improvement in quality as measured by determination index, the non-linear regression with Equation 11 was not considered further.

3.0 DISCUSSION

3.1 Duration of Experiment

The two stage process for the biochemical consumption of oxygen is supported by our results, yielding close correspondence between observed and calculated results. The failure of the non-linear regression to yield meaningful values for samples 6, 7, 8 and 9 for formula 9 ascribed to the two stage process represents a possible shortcoming in preparing a simple protocol for data handling. One potential reason for this non-convergence in certain sets of BOD data is early termination of the experiment. Thus the contribution of the slow stage may not be clearly discerned because of the contribution of the fast term and experimental scatter. In other words, if the LT-BOD study had been continued, convergent results would probably have been obtained.

A verifiable corollary of this statement is that if data sets yielding convergent two term solutions had been cut short, results would not have been satisfactory. This was tested by re-evaluating data sets but using BOD results only up to days 150, 120, 90, 70 and 50. The results noted in Table 6 clearly indicate a decrease in the experimental term leads to fewer satisfactory fits among the data sets.

The pattern emerging is that for shorter term experiments, non-linear regression yields non-convergent or obviously unsatisfactory solutions for two term exponential fits. As still shorter data sets are employed, the regression yields single term exponentials as best fits.

TABLE 6

Qualitative Results of Non-Linear Regression on Limited Data Sets

Sample	BOD Data Employed up to Day:						
	200+	150	144	120	90	70	50
1	-	-	S	S	S	S	S
2	-	-	S	1	S	2	NC
3	-	-	S	2	S	NC	NC
4	-	-	S	S	2	NC	NC
5	-	-	S	S	S	2	1
6	-	-	NC	1	1	1	2
7	-	-	NC	NC	NC	1	1
8	-	-	NC	NC	1	1	1
9	-	-	NC	NC	NC	1	1
10	-	-	S	2	NC	NC	1
11	S	S	S	S	2	S	2
12	S	S	S	2	2	2	1
13	S	S	S	S	S	S	NC
14	S	S	S	S	S	2	2
15	S	S	S	2	NC	NC	1
16	S	S	S	S	S	NC	1
% Satisfactory	100	100	75	44	44	19	6

S - Satisfactory solution to two term exponential fit

NC - No convergence for two term exponential fit (regression program failed to meet termination criteria in 300 iterations)

2 - Unsatisfactory solution to two term exponential fit (UBOD at shorter terms exceeded 10% deviation from longest term UBOD)

1 - Fit defaulted to single term exponential

TABLE 7

Non-Linear Regression Results on Sample 13 for Decreasing Data Sets

A. Solutions for the Two Term Expression, Equation 9

Duration (days)	Total UBOD	$UBOD_1$	k_1	$UBOD_2$	k_2	Determination Index
217	2154	640.1	0.126	1514	0.0256	0.9989
150	2129	501.5	0.158	1627	0.0285	0.9991
120	2121	458.5	0.171	1658	0.0297	0.9990
90	2098	409.4	0.189	1689	0.0312	0.9989
70	2198	544.6	0.153	1653	0.0258	0.9987
50*	No convergence					

B. Solutions for the One Term Expression, Equation 4

Duration	UBOD	k	Determination Index
217	2111	0.0377	0.9873
150	2068	0.0401	0.9903
120	2035	0.0420	0.9907
90	1980	0.0451	0.9920
70	1927	0.0481	0.9918
50	1794	0.0559	0.9930

TABLE 8

Non-Linear Regression Results on Sample 16 for Decreasing Data Sets

A. Solutions for the Two Term Expression, Equation 9

Duration (Days)	Total UBOD	$UBOD_1$	k_1	$UBOD_2$	k_2	Determination Index
217	2575	1407	0.122	1168	0.0193	0.9913
150	2635	1522	0.114	1113	0.0157	0.9927
120	2705	1581	0.110	1124	0.0132	0.9916
90	2620	1516	0.114	1104	0.0161	0.9900
70	No convergence					
50	No convergence					

B. Solutions for the One Term Expression, Equation 4

Duration	UBOD	k	Determination Index
217	2466	0.0498	0.9515
150	2400	0.0548	0.9578
120	2340	0.0596	0.9649
90	2257	0.0669	0.9766
70	2185	0.0735	0.9797
50	2069	0.0849	0.9878

Another aspect of the changing data set is the quality of the fits as measured by the determination index. The detailed results of Samples 13 and 16 are noted in Tables 7 and 8. The UBOD's are illustrated in Figures 15 and 16. Thus as shorter BOD data sets are employed, the correlation index for the two term fit changed little whereas the index for the single term fit improved for Sample 13 from 0.9873 to 0.9930 as the data base decreased from 217 to 50 days. Simultaneously, however, the UBOD declined by 15%. Similar trends can be see for Sample 16. Thus the apparent improvement in the single term exponential equation linear regression results is not necessarily desirable, as the improvement is actually due to a loss of information. This reflects the greater relative contribution of the fast portion of the UBOD in the early part of the BOD study.

Therefore the single term fit yields UBOD's within about 10% of the two term fit for data sets of over 200 days, but diverges more and more as the data set is restricted to shorter times. The two term solution remains fairly consistent until below 150 days of data is employed. At that point, non-linear regression starts to yield either non-convergent, single-term, or unsatisfactory (more than 10% deviation from longest duration result) fits.

The time required to consume half of the slow portion of the UBOD - the half-life - can also give an indication of the necessary duration of the experiment. Thus to utilize one half of the slow portion of the UBOD, hereafter termed $UBOD_2$:

$$\begin{aligned} BOD_2 \ consumed &= 0.5 \ UBOD_2 \\ &= UBOD_2 \ (1 - e^{-kt}) \end{aligned} \qquad (13)$$

Thus

$$UBOD_2 \ (1 - e^{-kt}) = 0.5 \ UBOD_2 \qquad (14)$$

or

$$e^{-kt} = 0.5 \qquad (15)$$

FIGURE 15: COMPARISON OF 1 AND 2 TERM UBODS FOUND FOR SAMPLE 13

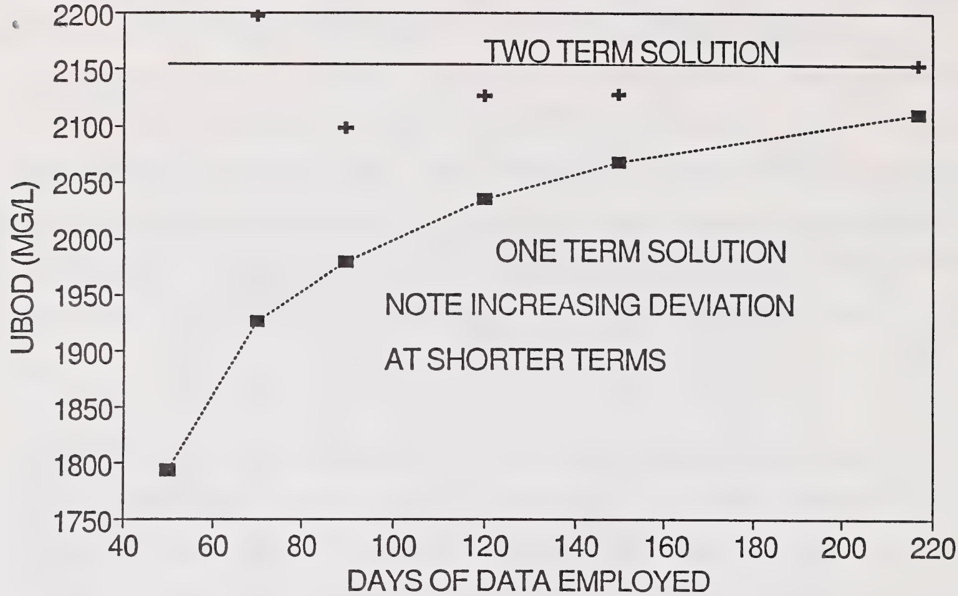
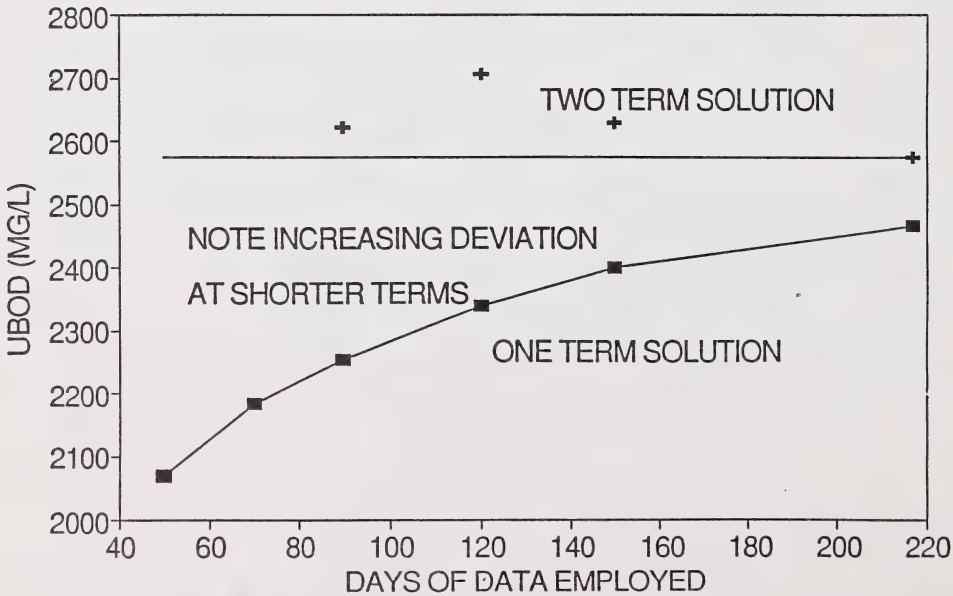


FIGURE 16: COMPARISON OF 1 AND 2 TERM UBODS FOUND FOR SAMPLE 16



By taking natural logarithms and solving for t:

$$t = 0.693 \; / \; k \tag{16}$$

The rates of the $UBOD_2$ noted in Table 3 for convergent solutions ranged from 0.0065 to 0.0279, however a geometric mean of 0.0145 was observed. For such a rate constant a half life of 48 days would be observed. Extension of the experiments to two or three half lives to allow expression of either 75% or 87.5% of $UBOD_2$, would require 96 or 144 days respectively, based on this mean. The fortuitous finding that 144 days is both three half lives, as well as the shortest duration noted in Table 6 that yielded over 50% satisfactory non-linear regression results, reinforces the point that reliable data analysis requires at least this duration.

This agrees well with a study by Fenske (20), in which he concluded that UBOD analyses should be run at least 80 days, and that the two term equation was superior to the single term equation. Our study suggests durations of up to 200 days give more reliable results.

3.2 Confidence Limits

The failure of non-convergent data sets to yield reasonable values although usually giving typical determination indices raises the need to examine the confidence that can be placed in the regression results. Preliminary evaluation of aspects of confidence limits was carried out, based on the work of Marske and Polkowski (21).

These workers focussed on the 95% confidence limit of solutions to the one term exponential expression, Equation 4. To determine the best fit to Equation 4, which will yield a unique value for UBOD and k, the unexplained sum of squares (USS) must be minimized. However to determine a confidence region, a critical USS not quite as demanding as the minimum USS must be determined. Then all combinations of UBOD and k for Equation 4 that do not exceed this critical USS are satisfactory. In practical terms, this will yield a satisfactory region on a plot of UBOD against k. This critical USS (USS_C) can be approximated as follows:

$$USS_C = USS\left[1 + \left(\frac{p}{n - p}\right) F\alpha, (p, n - p)\right] \tag{17}$$

where p = number of parameters estimated

n = number of observations

Fα = α significance point of the F distribution with p and n - p degrees of freedom.

For example, selection of a 95% confidence limit for sample 3 would yield:

$$USS_C = 60409 = 57922\ [1 + (2/143)\ 3.07] \tag{18}$$

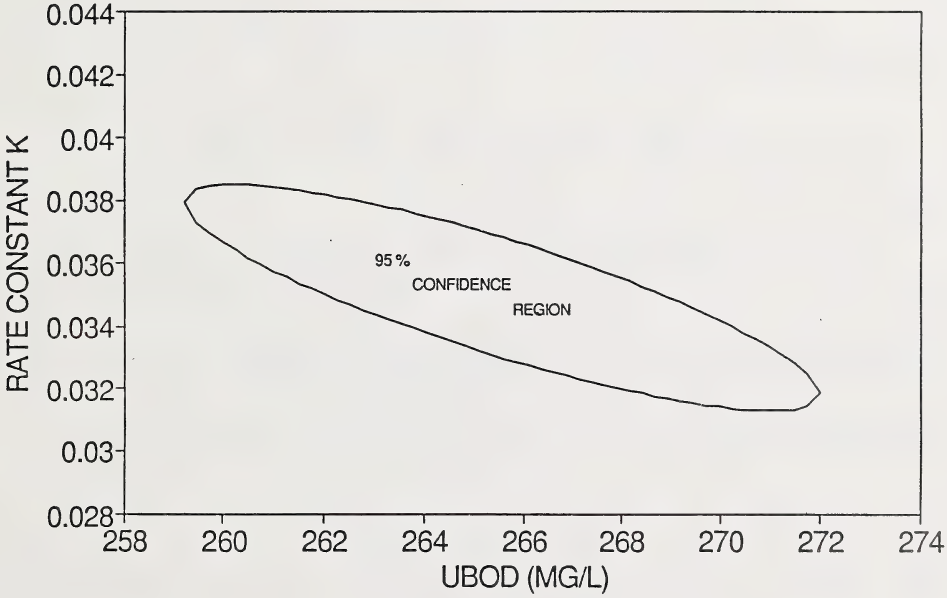
where 3.07 is the 5% significance point of the F distribution with 2 and 143 degrees of freedom. This is illustrated in Figure 17. All of the area inside the line is the 95% confidence region.

In such a two dimensional plot, a desirable result would be a small region centred on the least squares best fit parameters. Conversely a poor result would resemble a long valley, with a wide variety of possible solutions to UBOD and k within the 95% confidence region. If the valley was long but narrow, it could be described as a 95% confidence line on the two dimensional surface.

The situation for the two term exponential equation under consideration here is more complex. As four variables are involved, a confidence region within a 4 dimensional space must be evaluated. Depending on the quality of the result, the confidence region could resemble a point, a line (one dimension), a surface (two dimensions), or a volume (three dimensions) within the 4 dimensional space.

A simplified approach was taken initially to inspect one of the non-convergent solutions. Thus Sample 6 was selected, and the fast UBOD term fixed at its value as found in Table 3. The resulting equation:

FIGURE 17: 95% CONFIDENCE REGION FOR THE ONE TERM SOLUTION FOR SAMPLE 3



$$BOD(t) = 320\ (1 - e^{-0.034\,t}) + UBOD_2\ (1 - e^{-k_2 t}) \tag{19}$$

was evaluated to determine the 95% confidence region. This region is illustrated in Figure 18. Essentially, any value from 162 mg/L and up can, with appropriate choice of k_2, meet the requirements of the 95% confidence limits. The slow BOD parameters are therefore poorly defined in this case.

A similar procedure was followed for Sample 13, and the result illustrated in Figure 19. In contrast to the previous example, the 95% confidence region is a small area around the best fit value, which can be described as:

$$UBOD_2 = 1514 \pm 4\ ;\ k_2 = 0.0256 \pm 0.0002$$

A clear difference has appeared between the non-convergent and convergent non-linear regression results. The non-convergent "solution" confidence region extends from what could be considered reasonable to an area of very high $UBOD_2$ and low k_2.

3.3 Rate Constants

To clearly define what is "reasonable", the characteristics of the fast and slow rates were investigated, both for our data and other non-linear regression work on treated effluents reported in the literature. The results are summarized in Table 9. The ratio between k_1 and k_2 varies from 4.1 to 28.1. The geometric mean of the ratio derived from this study is 8.9, matching fairly closely the geometric mean of the literature work, 12.6. For all data in the table, the geometric mean is 10.8.

FIGURE 18: 95 % CONFIDENCE REGION FOR SAMPLE 6 OF SLOW UBOD AND K

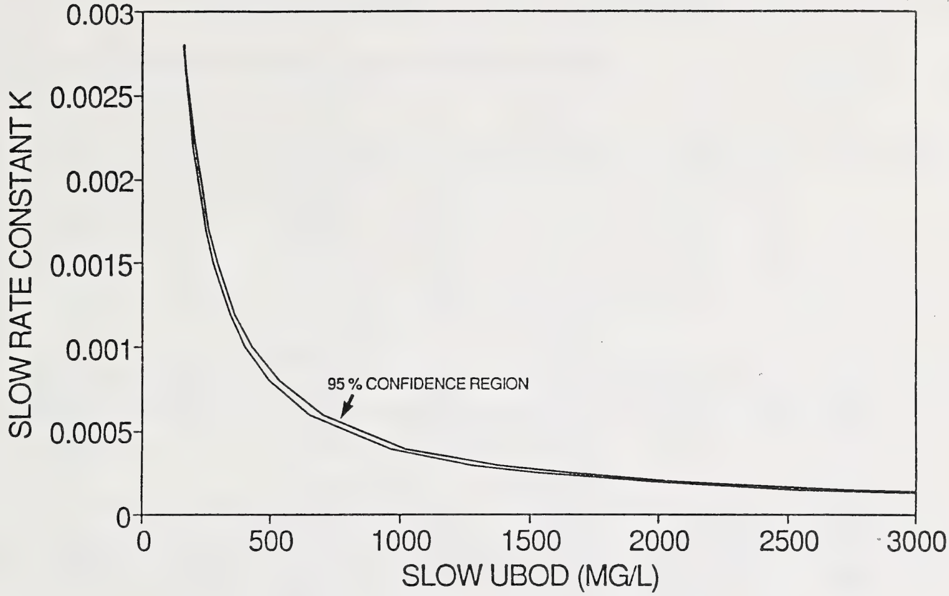


FIGURE 19: 95 % CONFIDENCE REGION FOR SAMPLE 13 OF SLOW UBOD AND K

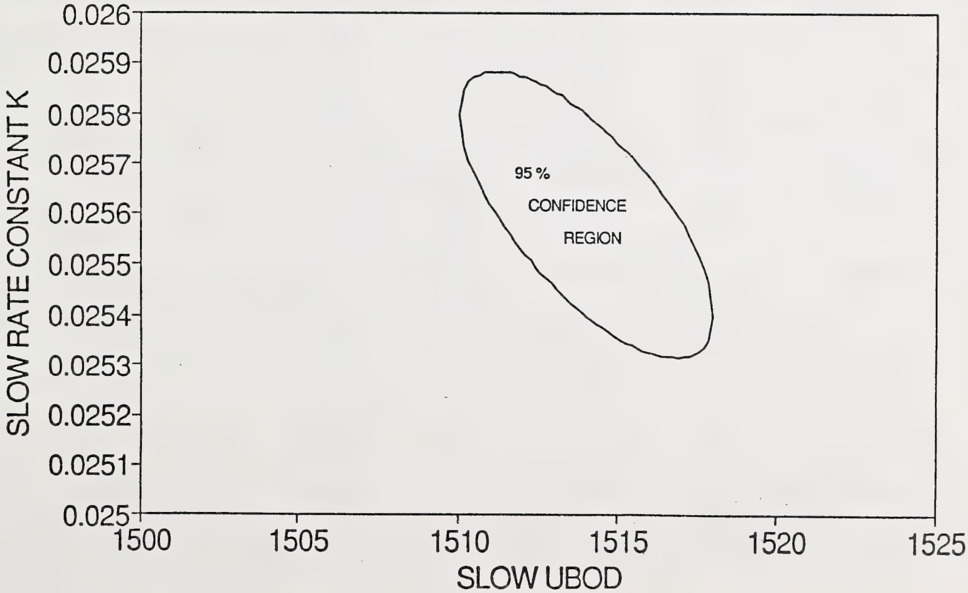


TABLE 9

Comparison of Fast and Slow Rate Constants for This and Other Studies

Sample	k_1	k_2	Ratio (k_1/k_2)
1	0.299	0.0279	10.7
2	0.312	0.0258	12.1
3	0.118	0.0065	18.2
4	0.142	0.0142	10.0
5	0.068	0.0108	6.3
10	0.130	0.0065	20.0
11	0.130	0.0109	11.9
12	0.060	0.0083	7.2
13	0.126	0.0256	4.9
14	0.096	0.0216	4.4
15	0.130	0.0194	6.7
16	0.122	0.0193	6.3
Ref 4	0.400	0.017	23.5
Ref 5	0.420	0.085	4.9
Ref 13	0.478	0.0183	26.1
Ref 13	0.358	0.0871	4.1
Ref 13	0.0903	0.0207	4.4
Ref 13	0.281	0.0246	11.4
Ref 13	0.252	0.0142	17.7
Ref 13	0.428	0.0172	24.9
Ref 13	0.480	0.0171	28.1
Ref 13	0.450	0.0172	26.2
Ref 15	0.455	0.072	6.3
Ref 20	0.067	0.009	7.4
Ref 20	0.25	0.016	15.6
Ref 20	0.16	0.007	22.9
Ref 20	0.41	0.041	10.0
Geometric Mean	0.202	0.0187	10.8
95% Confidence Limits	0.151 - 0.270	0.014 - 0.025	8.3 - 14.1

Determination of the confidence limits of the data according to the method of Sokal et al (31) indicates k_2 can be expected to be between one quarter and one thirtieth of k_1. Thus either the non-linear regression can be terminated before the ratio k_1/k_2 exceeds a given value, or a forced fit for a chosen ratio of k_1/k_2 could be undertaken.

The results of early termination of the iterative program for samples 6, 7, 8 and 9 are noted in Table 10.

TABLE 10

Ultimate Biochemical Oxygen Demand and Rate Constants for Blank-Corrected BOD Results Determined by Non-Linear Regression with Early Termination of Iterative Program

Sample	Total UBOD	$UBOD_1$	k_1	$UBOD_2$	k_2	Determination Index	Single Term UBOD*
6	487	309	0.0347	178	0.00324	0.9954	373
7	340	178	0.0712	261	0.00408	0.9872	276
8	445	211	0.0593	234	0.00493	0.9760	315
9	231	75	0.159	156	0.00748	0.8000	176

* Results from Table 2

With the exception of Sample 9, the determination index closely matched the previous value noted in Table 3 and reproduced here in Table 11. Sample 6, 7 and 8 yield results within or very close to the 95% confidence region. Similarly, the fast UBOD and rate closely resembled the original values determined after 300 iterations of the non-linear regression program. Thus even for non-convergent solutions, the fast UBOD and k_1 are relatively stable.

TABLE 11

Fast UBOD and Rate Constants for 300 Iteration and Early Termination Results

Sample	300 Iteration (Table 3)			Early Termination (Table 10)		
	$UBOD_1$	k_1	Determination Index	$UBOD_1$	k_1	Determination Index
6	309	0.035	0.9954	320	0.034	0.9955
7	178	0.071	0.9872	192	0.067	0.9880
8	211	0.059	0.9760	229	0.056	0.9775
9	75	0.159	0.8000	80	0.252	0.8498

How sensitive is a total UBOD value determined for a supposedly problem-free data set? A simple test was carried out employing the UBOD data for sample 13, but forcing the ratio of k_1/k_2 to values of 5, 10 and 20:1. The best fits for both the unconstrained non-linear regression and these forced examples are noted in Table 12. Clearly, manipulation of the rates has a dramatic effect on the fast $UBOD_1$, and a proportionally smaller effect on the slow $UBOD_2$. The determination index dropped slightly from 0.9989 to 0.9968, still a good fit. Comparison of the USS of the results suggests the best fit is three times better than the poorest fit, which was the 20:1 ratio. However the total UBOD varied by less than 1% across the full range of trials. Therefore the total UBOD is not sensitive to the perturbations introduced here.

A possible corollary of this conclusion is that $UBOD_1$ and $UBOD_2$ are very sensitive to minor variations in the database, whereas the total UBOD is resistant to such effects. This can be tested by examination of duplicate LT-BOD tests to assess correlation of $UBOD_1$, $UBOD_2$ and total UBOD values determined. To reduce experimental variations, the most reliable $UBOD_1$, $UBOD_2$, k_1 and k_2 results would be obtained from LT-BOD trials of very extended duration.

TABLE 12

Best Fits for Sample 13 if k_1 and k_2 Maintained at Various Ratios

Ratio k_1/k_2	Total UBOD	$UBOD_1$	k_1	$UBOD_2$	k_2	Determination Index
4.9*	2154	640	0.126	1514	0.0256	0.9989
5	2150	597	0.132	1553	0.0264	0.9989
10	2142	391	0.292	1751	0.0292	0.9980
20	2135	304	0.617	1831	0.0309	0.9968

* Unconstrained result from Table 3.

3.4 Summation

This manuscript reviews and integrates the various techniques of measurement and calculation required to arrive at valid UBOD estimates in environmental waters. It builds from a literature base and investigates various curve fit models from which the resultant UBOD number can be "reliably" estimated. An attempt was made to apply statistical criteria to the choice of a preferred model and to replace subjectivity with objectivity. These effects led to the following novel aspects:

1. Analyses of the effects of experimental duration proved the necessity for extended terms both to achieve successful analysis for the two term model and accurate results for the one-term model.

2. Evaluation of the reliability of the results purely by statistical means (determination coefficient) was shown to be misleading. Thus results from a one-term model of very limited duration had a similar determination coefficient to a successful two term model, although the UBOD was very different. Furthermore, the one-term model yielded statistically poorer results as the data base was expanded.

3. Analysis of available data indicated an average ratio between the fast and slow rate constants of 10:1.

4. Sensitivity analysis was conducted on the two-term model to preset ratios of reaction rates, and showed stable total UBOD values.

4.0 CONCLUSIONS

Primary Conclusions

1. Both evaluation of the residuals between predicted and observed data and the determination indices of the non-linear regression results of one and two term models indicate a successful two term regression result is definitively superior to a one term fit.

2. One cause of non-convergent results during non-linear regression with the two-term expression is early termination of the experiment. Thus less than half of the samples yielded satisfactory results when 120 days data was employed. Terms approaching 200 days are recommended.

3. A preliminary test of sensitivity of the two term solution to preset ratios of k_1 to k_2 showed relative instability in the fast and slow UBOD values, however the total UBOD remained very stable.

Secondary Conclusions

4. Non-linear regression analysis on blank corrected BOD data yielded UBOD values for all samples tested with a single term exponential equation and most samples tested with a two term exponential expression.

5. For data sets of successively shorter duration, the results for the two term expression remains fairly consistent until it fails completely. The results for the single term expression fall farther and farther below the best result, but show an apparent improvement in determination index. This apparent improvement is actually due to a loss of information of the long term characteristics of the BOD curve.

6. The 95% confidence limits were generated and displayed graphically for a two dimensional situation. Specific application of this concept to one of the non-convergent sample results showed a highly unstable solution, whereas a sample yielding a convergent result produced confidence limits with less than 1% maximum deviation from the best fit values.

7. The ratio of the fast and slow rate constants, k_1 and k_2, for convergent results in this and other studies cover a range from 4.1 to 28.1. Termination of the iterative non-linear regression program before the unsatisfactory non-convergent results exceed the ratio yield solutions which have UBOD's exhibiting greater similarity to the single-term UBOD values.

8. A conservative (maximum) UBOD can be estimated by taking 1.5 times the single term UBOD for databases of at least 120 days duration.

REFERENCES

1. APHA, AWWA, WPCF, "Standard Methods for the Examination of Water and Wastewater" Seventeenth Edition, 1989. Section 5210, p 5-3.
2. APHA, AWWA, WPCF, "Standard Methods for the Examination of Water and Wastewater" Fourteenth Edition, 1975. Section 507, p 548.
3. Stamer, J.K., McKenzie, S.W., Cherry, R.N., Scott, C. T., and Stamer, S.L., J. Water Pollution Control Fed., 1979, 51, 918.
4. McKeown, J.J., Brown, L.C., and Martone, C.H., Wat. Sci. Tech., 1981, 13, 363.
5. Hiidenheimo, H. and Wilson, M.F., Water Res., 1974, 8, 363.
6. Wisconsin State Laboratory of Hygiene, "Oxygen Demand, Biochemical, Long-Term, Method 260.2", 1981.
7. Whittemore, R., "Ultimate Biochemical Oxygen Demand", submitted to Standard Methods Editorial Board, 1988.
8. Woznow, D., Hill, H., Skinner, F., and van Roodselaar, A., "Oxygen Demand, Ultimate Biochemical, Provisional Method", Alberta Environmental Centre, Vegreville, Alberta, Canada, TOB 4LO, 1989.
9. Skinner, F. and van Roodselaar, A., "Nutrient Effects in Long-Term Biochemical Oxygen Demand Studies", in preparation.
10. Skinner, F., and van Roodselaar, A., "Ultimate Biochemical Oxygen Demand: Effects of Environmental Factors", in preparation.
11. Thomas, H.A., Sewage Works Journal, 1937, 9, 425.
12. Monod, J., "La Croissance des Culture Bacteriennes", Paris, Harmann, 1942.
13. Whittemore, R.C., and Hovis, J., "A Review of Ultimate BOD Estimation and Its Kinetic Formulation for Pulp and Paper Mill Effluents", NCASI Technical Bulletin No. 382, 260 Madison Avenue, NY, NY, 10016, October 1982.
14. Whittemore, R.C., "A Review of the Separation of Carbonaceous and Nitrogenous BOD in Long-term BOD Measurements", NCASI Technical Bulletin No 461, 260 Madison Avenue, NY, NY, 10016, May 1985.
15. Raabe, E.W., J. Water Pollution Control Fed., 1968, 40, Supplement 5, R145.

16. Crawford, D.L., Floyd, S., Pometo III, A.L., and Crawford, R.L., Can. J. Microbiol. 1977, 23, 434.
17. Kirk, T.K. and Farrell, R.L., Ann. Rev. Microbiol, 1987, 41, 465.
18. Bouveng, H.O. and Solyom, P. Svensk Paperstidn., 1973, 76, 26.
19. Hwang, J.H., "User's Manual for Parameter Estimation For First Order Ultimate BOD Decay, BODFO", NCASI Technical Publication No. 529, 260 Madison Avenue, NY, NY, 10016, July 1987.
20. Fenske, B.A. American Water Resources Association, Wisconsin Section, 6th Annual Meeting, March 4-5, 1982, Stevens Point, Wisconsin.
21. Marske, D.M. and Polkowski, L.B., J. Water Pollution Control Fed., 1972, 44, 1987.
22. Leduc, R., Unny, T.E., and McBean, E.A., Water Res., 1986, 20, 625.
23. Constable, T.W. and McBean, E.A., Can. J. Civ. Eng., 1977, 4, 371.
24. Constable, T.W. and McBean, E.A. Can. J. Civ. Eng., 1977, 4, 462.
25. Berthouex, P.M., Asce, A.M., and Hunter, W.G., J. Sanitary Engineer. Div., ASCE, 1971, 97, 8202.
26. Brown, H.B. and Pico, R.F., Proceedings of the 34th Industrial Waste Conference, May 8-10, 1979, Page 326.
27. Hewitt, J., Hunter, J.V. and Lockwood, D., Water Res., 1979, 13, 325.
28. Fitall Research Edition 4.01, MTR Software, 1988, P.O. Box 902, Station P, Toronto, Ontario, M5S 2Z2.
29. Young, R.H.F., Ryckman, D.W. and Buzzell, J.C., J. Water Pollution Control Fed., 1968, 40, R354.
30. Hughes, A. and Grawoig, D., "Statistics: A Foundation for Analysis", p 386 ff, Addison-Wesley Publishing Co., Reading, MA, 1971.
31. Sokal, R.R. and Rohlf, F.J., "Biometry", second edition, p 419 ff, W.H. Freeman and Co, New York, New York, 1981.

APPENDIX

Procedure to Prepare LT-BOD Data for Non-Linear Regression

The LT-BOD study yields an extensive list of 60 to 100 DO readings and 5 - 20 reaeration readings for each sample. These must be manipulated to yield a cumulative DO demand for the course of the experiment. Then, in conjunction with observed blank data and available nitrate results, a LT-BOD database corrected for the contribution of added nutrients is prepared. This database, after suitable weighting, is analyzed by non-linear regression using both the one term and two term exponential equations. The procedure is illustrated in the flow chart (Figure A-1).

Cumulative BOD Values

For DO readings from the initiation of the study to the first aeration, the difference between the initial DO value, DO (o), and the observed daily reading, DO (t), yields the cumulative DO depletion:

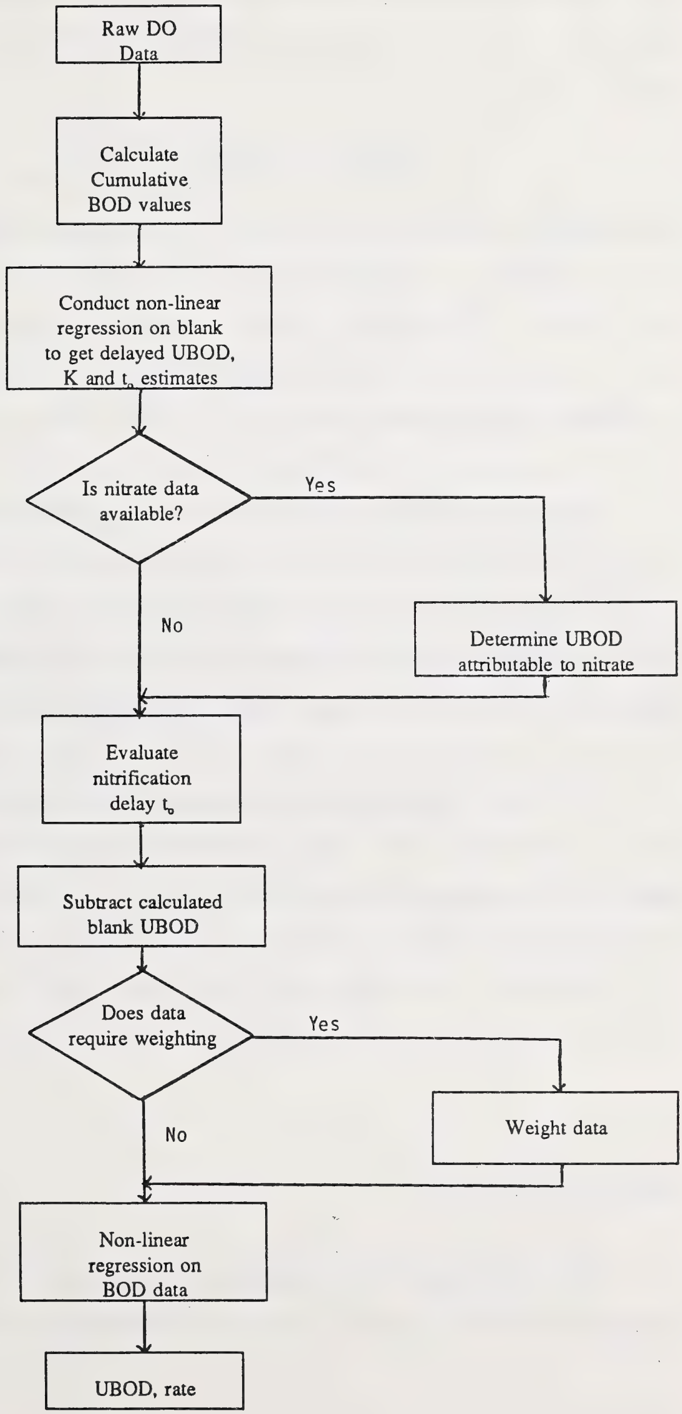
$$Cumulative\ DO(t) = DO(o) - DO(t) \qquad (A1)$$

When reaeration is required, a final reading before reaeration must be obtained to yield cumulative DO up to that point, here termed Σ(R1). After reaeration, a new initial DO reading DO (R1) must be taken and the sequence begins again. The cumulative value is now:

$$Cumulative\ DO(t) = \sum(R1) + DO(R1) - DO(t) \qquad (A2)$$

All cumulative DO values must finally be divided by the concentration of sample in the BOD test to yield the BOD values.

Figure A-1. Derivation of UBOD and Rate From Raw DO Data



Blank Correction

The primary source of DO variation in the blank is attributable to the effects of nitrification. The delayed exponential equation discussed in the body of the paper and used to describe nitrification is:

$$NBOD(t) = UBOD_3\ (1 - e^{-k(t-t_o)}) \qquad (A3)$$

where NBOD is nitrogenous biochemical oxygen demand, $UBOD_3$ is the UBOD attributable to the delayed BOD, k is the respective rate, and t_o is the delay before nitrification begins. These three variables, $UBOD_3$, k, and t_o, must be determined to yield a satisfactory description of the blank DO requirements. The DO demand data from the nutrients containing blank is subjected to non-linear regression using Equation A3 to yield values for these three variables.

These values can be considered preliminary estimates for purposes of blank correction. However the experimental design may allow refinement of the value of $UBOD_3$. Thus if nitrate analyses were conducted at the initiation and termination of the experiment, the oxygen required to account for the observed increase in nitrate can be employed as $UBOD_3$. The oxygen consumed corresponds to 4.57 times the observed increase in nitrate nitrogen.

Finally the delay of onset of nitrification in the sample under study may differ from the blank. As discussed in the body of the report, modification of the delay time and visual evaluation of the corrected curve represents one method of adjustment. Alternatively, the delay determined for the blank by non-linear regression could be employed without modification, and the corrected data in the BOD curve in the time interval near the initiation of nitrification, typically 15 - 35 days, not included in the future analysis by giving it a low weighting.

Weighting of BOD Data

After satisfactory blank parameters are obtained, the calculated blank BOD values are subtracted from the sample BOD values. The weighting of different portions of the BOD data

must now be considered. Daily BOD determinations are available for the first 30 or 40 days. However near the end of the study, BOD values are typically obtained at weekly intervals. Without appropriate time-weighting, non-linear regression on the BOD data would give undeserved prominence to the initial phase of fast BOD expression.

The method used in this study was to interpolate BOD values for any days on which readings were not taken. Then each day achieves an equal weighting. Some non-linear regression programs allow a higher weighting to be assigned to data obtained during times of infrequent analysis.

Determination of UBOD and Rate

The corrected and weighted BOD data is now subjected to non-linear regression. The best values determined for Equations 4 and 9 represent the respective UBOD's and rates.

Not all iterative programs converge to the best possible result, termed a global minimum; some instead yield a local minimum. Thus reinitiation of the non-linear regression program with different estimates may yield significantly different values, the quality of the result being judged by the variance or the determination index. This step requires some experience on the part of the analyst, especially for the two term exponential expression, Equation 9.

Upon discovery of the global minimum for UBOD and rate, the process is complete. The UBOD and rate data must be reviewed in conjunction with related data as described in the body of the report.

Printed on Recycled Paper